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## THE INFLUENCE OF THE INGESTION OF DEAD TUBERCLE BACILLI UPON INFECTION.\*†

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DEAD tubercle bacilli, when injected into a guinea-pig or other animal, are capable of producing either local or general effects, depending chiefly on the number introduced. As to whether the ingestion of dead tubercle bacilli is harmful or beneficial, or without effect, the evidence is not clear, as there has been but little experimental work done on this subject.

In Germany and other European countries meat of tuberculous animals is not destroyed, as is required by the federal regulations of this country. Such meat in certain European countries is cooked under official surveillance and sold at a lower price. This is the so-called "Freibank system."

The question assumes great practical importance on account of the use of pasteurized milk, cooked meat from tuberculous animals, and other food products containing dead tubercle bacilli. It is of the utmost importance to determine if the susceptibility to tuberculosis is increased, decreased, or not altered by the previous eating of food containing tubercle bacilli killed by heat. The experiments which we wish to report were designed to simulate as closely as possible the effect produced by the ingestion of pasteurized milk or milk products.

The tubercle bacilli were heated at 60° C. for 30 minutes, mixed with butter, and fed to guinea-pigs daily (excepting Sundays) for 60 days. A low degree of heat was used to prevent altering the bacilli as much as possible and closely to simulate pasteurization. The animals were always given the dead bacilli mixed in the butter before receiving their usual morning feed. Control inoculations of the

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heated bacilli, made into the peritoneal cavity of five guinea-pigs, showed that the tubercle bacilli were dead.

In all these experiments the tubercle bacilli were fed as follows: The animal's mouth was opened and a little pellet of butter containing the tubercle bacilli placed upon the tongue. The animal was then released and it at once swallowed the material thus placed in the mouth. In placing the material upon the tongue it is convenient to use a piece of glass tubing into which the butter is pressed and can be squeezed out with a glass rod acting as a piston at the desired moment. In this manner animals may be fed without danger of inflicting wounds.

The butter used in these experiments was made in the laboratory from a specially good cream. The cream was obtained from a herd of tuberculin-tested cows, but was nevertheless first pasteurized to insure its freedom from live tubercle bacilli.

The culture used in these experiments is known as "Bovine Culture H," and was obtained from Dr. Leonard Pearson. It was isolated July 5, 1889, from the omentum of a guinea-pig which had been inoculated with an emulsion of caseous material from the mesenteric gland of a Jersey cow seven years old. The culture was grown on blood serum for 22 generations, then transplanted on glycerin-agar; it was either the 33d or 34th generation on glycerin-agar, and was virulent for guinea-pigs, rabbits, and cattle.

A four-weeks-old glycerin-potato culture was scraped off, suspended in water, and heated at 60° C. for 30 minutes; the bacilli were allowed to sediment and the supernatant water removed. The mass of dead tubercle bacilli was mixed with the butter in about equal proportions and kept in the ice-chest. At the time of feeding this was largely diluted with sterile butter and each guinea-pig given about 0.1 gm. of the mixture. Microscopic smears showed tubercle bacilli in each field lying singly or in groups.

The live bacilli used to infect the animals were taken from a 43-days-old glycerin-broth culture grown in six Fernbach flasks. The bacilli were dried at 37° C. and 100 gm. of the dried tubercle bacilli were mixed with an equal volume of butter. Each guinea-pig received about 0.1 gm. of the mixture, which was fed by the same method as described for the dead bacilli.

The details of the results are seen in the tables.

TABLE I.

DEAD TUBERCLE BACILLI FED DAILY EXCEPT SUNDAYS FOR 60 DAYS PRIOR TO ONE FEEDING OF LIVE TUBERCLE BACILLI.

No. of Guinea-Pig	Day of Death	Initial Weight in Grams	Result	TUBERCULOSIS IN GLANDS										Remarks	
				Submental	Submaxillary	Cervical	Axillary	Inguinal	Bronchial	Anterior mediastinal	Retro-peritoneal	Sublumbar	Retro-hepatic		Mesenteric
1	80	300	Generalized tuberculosis	+	+	+	+	+	+	+	o	+	+	+	All had tuberculosis of liver, lungs, and spleen
2	92	275	"	+	+	+	+	+	+	+	+	+	+	+	
3	110	275	"	+	+	+	+	+	+	+	+	+	+	+	
4	112	260	"	+	+	+	+	+	+	+	+	+	+	+	
5	118	300	"	+	+	+	+	+	+	+	+	+	+	+	
6	120	270	"	+	+	+	+	+	+	+	+	+	+	+	
7	138	330	"	+	+	+	+	+	+	+	+	+	+	+	
8	142	340	"	+	+	+	+	+	+	+	+	+	+	+	
9	148	275	"	+	+	+	+	+	+	+	+	+	+	+	
10	155	260	"	+	+	+	+	+	+	+	+	+	+	+	
11	70	250	"	+	+	+	+	+	+	+	+	+	+	+	
12	71	320	"	+	+	+	+	+	+	+	+	+	+	+	
13	147	270	"	+	+	+	+	+	+	+	+	+	+	+	
14	112	300	"	+	+	+	+	+	+	+	+	+	+	+	
15	93	370	"	+	+	+	+	+	+	+	+	+	+	+	
16	71	290	"	+	+	+	+	+	+	+	+	+	+	+	
17	71	275	"	+	+	+	+	+	+	+	+	+	+	+	
18	71	245	"	+	+	+	+	+	+	+	+	+	+	+	
19	70	275	"	+	+	+	+	+	+	+	+	+	+	+	
20	79	290	"	+	+	+	+	+	+	+	+	+	+	+	
21	79	310	"	+	+	+	+	+	+	+	+	+	+	+	
22	79	310	"	+	+	+	+	+	+	+	+	+	+	+	
23	77	290	"	+	+	+	+	+	+	+	+	+	+	+	
24	98	330	"	+	+	+	+	+	+	+	+	+	+	+	
25	105	290	"	+	+	+	+	+	+	+	+	+	+	+	
26	92	260	"	+	+	+	+	+	+	+	+	+	+	+	
27	107	290	"	+	+	+	+	+	+	+	+	+	+	+	
28	111	260	"	+	+	+	+	+	+	+	+	+	+	+	
29	113	275	"	+	+	+	+	+	+	+	+	+	+	+	
30	117	240	"	+	+	+	+	+	+	+	+	+	+	+	
31	117	275	"	+	+	+	+	+	+	+	+	+	+	+	
32	176	275	"	+	+	+	+	+	+	+	+	+	+	+	
33	110	275	"	+	+	+	+	+	+	+	+	+	+	+	
34	151	290	"	+	+	+	+	+	+	+	+	+	+	+	
35	151	285	"	+	+	+	+	+	+	+	+	+	+	+	
36	151	300	"	+	+	+	+	+	+	+	+	+	+	+	
37	127	275	"	+	+	+	+	+	+	+	+	+	+	+	
38	185	355	"	+	+	+	+	+	+	+	+	+	+	+	
39	151	315	"	+	+	+	+	+	+	+	+	+	+	+	
40	130	270	"	+	+	+	+	+	+	+	+	+	+	+	
41	120	285	"	+	+	+	+	+	+	+	+	+	+	+	
42	126	300	"	+	+	+	+	+	+	+	+	+	+	+	
43	120	300	"	+	+	+	+	+	+	+	+	+	+	+	
44	120	260	"	+	+	+	+	+	+	+	+	+	+	+	
45	148	300	"	+	+	+	+	+	+	+	+	+	+	+	
46	140	275	"	+	+	+	+	+	+	+	+	+	+	+	
47	133	275	"	+	+	+	+	+	+	+	+	+	+	+	

Average 113.

+ = Tuberculosis; o = Normal; . = Not noted.

TABLE 2.  
 CONTROLS : ONE FEEDING OF LIVE TUBERCLE BACILLI.

No. of Guinea-Pig	Day of Death	Initial Weight in Grams	Result	TUBERCULOSIS IN GLANDS										Remarks	
				Submental	Submaxillary	Cervical	Axillary	Inguinal	Bronchial	Anterior mediastinal	Retro-peritoneal	Sublumbar	Retro-hepatic		Mesenteric
1	79	320	Generalized tuberculosis	o	o	o	o	+	+	o	o	+	+	+	All had tuberculosis of liver, lungs, and spleen
2	79	320	"	+	+	+	o	+	+	+	+	+	o	+	
3	79	275	"	+	+	+	+	+	+	+	+	+	+	+	
4	79	275	"	+	+	+	+	+	+	+	+	+	+	+	
5	71	260	"	+	+	+	+	+	+	+	+	+	+	+	
6	105	275	"	+	+	+	+	+	+	+	+	+	+	+	
7	80	230	"	+	+	+	+	+	+	+	+	+	+	+	
8	105	265	"	+	+	+	+	+	+	+	+	+	+	+	
9	101	300	"	+	+	+	+	+	+	+	o	o	o	o	
10 11 12 13 14 15 16 17 18 19 20 21 22	94 108 107 108 111 111 111 114 116 118 118 118	300 285 210 270 280 230 335 255 345 285 350 320 335	" " " " " " " " " " " "	+	+	+	o	o	+	+	+	+	+	+	Lesions above diaphragm. Ax. and ing. glands slightly enlarged
	+	+	+	o	o	+	+	+	+	+	+	+	+	+	
	+	+	+	o	o	+	+	+	+	+	+	+	+	+	
	+	+	+	o	o	+	+	+	+	+	+	+	+	+	
	+	+	+	o	o	+	+	+	+	+	+	+	+	+	
	+	+	+	o	o	+	+	+	+	+	+	+	+	+	
	+	+	+	o	o	+	+	+	+	+	+	+	+	+	
	+	+	+	o	o	+	+	+	+	+	+	+	+	+	
	+	+	+	o	o	+	+	+	+	+	+	+	+	+	
	+	+	+	o	o	+	+	+	+	+	+	+	+	+	
	+	+	+	o	o	+	+	+	+	+	+	+	+	+	
	+	+	+	o	o	+	+	+	+	+	+	+	+	+	
	+	+	+	o	o	+	+	+	+	+	+	+	+	+	
23 24 25 26	110 118 176 176	300 275 305 250	" " Normal Generalized tuberculosis	+	+	+	o	o	+	+	+	+	+	+	Perit. surface of colon studded with tubercles
	+	+	+	o	o	+	+	+	+	+	+	+	+	+	
	+	+	+	o	o	+	+	+	+	+	+	+	+	+	
	+	+	+	o	o	+	+	+	+	+	+	+	+	+	
	+	+	+	o	o	+	+	+	+	+	+	+	+	+	
27 28 29 30 31 32 33 34 35 36 37 38	172 157 128 128 126 122 123 122 146 142 140 120	335 300 255 310 285 295 285 235 270 270 290 270	" " " " " " " " " " "	+	+	+	o	o	+	+	+	+	+	+	Chloroformed
	+	+	+	o	o	+	+	+	+	+	+	+	+	+	
	+	+	+	o	+	+	+	+	+	+	+	+	+	+	
	+	+	+	+	+	+	+	+	+	+	+	+	+	+	
	+	+	+	+	+	+	+	+	+	+	+	+	+	+	
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	+	+	+	+	+	+	+	+	+	+	+	+	+	+	
	+	+	+	+	+	+	+	+	+	+	+	+	+	+	
	+	+	+	+	+	+	+	+	+	+	+	+	+	+	
	39 40 41	132 84 85	255 260 275	" "	+	+	+	o	+	+	o	+	+	+	
+		+	+	+	+	+	+	+	+	+	+	+	+	+	
+		+	+	+	+	+	+	+	+	+	+	+	+	+	
42 43 44 45	84 158 49 63	340 325 290 310	" " " "	+	+	+	.	+	+	+	+	+	+	+	Killed by accident
	+	+	+	+	+	+	+	+	+	+	+	+	+	+	
	+	+	+	+	+	+	+	+	+	+	+	+	+	+	
	+	+	+	+	+	+	+	+	+	+	+	+	+	+	
46	62	275	"	+	+	+	+	+	+	+	+	+	+	+	

Average 108.

+ = Tuberculosis; o = Normal; . = Not noted.

One hundred guinea pigs about six weeks old were used in these experiments. They were divided into two lots; the first lot, consisting of 50, was fed for 60 days on the heated tubercle bacilli; the second lot, also consisting of 50 guinea-pigs, which served as the controls, was kept under the same conditions, but did not receive the heated bacilli. The entire 100 at the end of 60 days were given one feeding of the 43-days-old live tubercle bacilli in butter. During the entire course of the experiments all the animals were kept under exactly similar conditions. As soon as an animal died it was carefully autopsied, especial note being made of the involvement of the lymphatic glands.

Under the conditions of the experiments it is evident that the feeding of the dead tubercle bacilli to the guinea-pigs did not alter their susceptibility. It seemed to have no evident effect upon the subsequent course of the disease. It is not safe to conclude from this that the ingestion of dead tubercle bacilli under all circumstances is a harmless procedure, although this is perhaps indicated.

As a result of this work several interesting observations were noted. One is, that none of the animals fed with the dead bacilli lost weight during the 60 days this process continued. It is also evident that if a guinea-pig be given a sufficient amount it may be infected by ingestion, for all the animals with one exception became infected. In one instance (Guinea-pig 9, Table 2) the lesions of ingestion tuberculosis were all above the diaphragm; this guinea-pig had three enlarged and caseous glands in the neck, the bronchial and mediastinal glands were tuberculous, and the lungs tuberculous; the liver, spleen, and other abdominal organs were normal.

It is also evident that it takes much longer for guinea-pigs to die of ingestion tuberculosis than when the same material is inoculated into the peritoneal cavity. Five controls, which had received the same material into the peritoneal cavity, died in 13, 15, 15, 17, and 37 days respectively, whereas the average from Table 1 was 113 days and from Table 2 it was 108 days.